THE TAXONOMY OF *ENTEROMORPHA* LINK, 1820, (CHLOROPHYCEAE) IN THE NETHERLANDS

II. The section Proliferae

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ABSTRACT. On the basis of 182 living samples from 31 different stations, five *Enteromorpha* species were distinguished and described for the Netherlands coast within the section *Proliferae* (Bliding's «Prolifera Group», here including *E. torta*), namely *E. simplex* (Vinogradova) nov. comb. nov. stat., *E. prolifera* (O.F. Müll.) J.Ag., *E. radiata* J.Ag., *E. torta* (Mert. in Jürg.) Reinb., and *E. ahlneriana* Bliding. Unialgal cultures were isolated from part of the samples in order to test the validity of the taxonomic criteria and to test the growth responses to varying salinities. The macroscopic morphology of the plants appeared to present the most distinctive differences between the species within the section. The morphology of the basal parts and the morphology and distribution of filiform branchlets, if present, offered additional criteria, whilst cell sizes and cell arrangements showed some, mostly minor differences. Diversity in the species of the section *Proliferae* in the Netherlands agrees well with diversity recognized until now. *E. simplex*, *E. prolifera* and *E. ahlneriana* are euryhaline species occurring in euhaline to mesohaline waters. *E. prolifera* occurs even in oligohaline waters. *E. simplex* cultures grow well at salinities of 9-34‰ S, *E. prolifera* and *E. ahlneriana* cultures even at 1.5-34‰ S. The less euryhaline species *E. torta* and *E. radiata* occur only on littoral mud and sandy mudflats, *E. torta* preferring the upper zones. In culture they grow well in salinities ranging from 9-34‰ S.

INTRODUCTION

The present study is the third of a series on the taxonomy of Ulvales in the Netherlands. BLIDING's (1963, 1968) revisions of European Ulvales have led to the distinction of many more species (and subspecific taxa) in *Enteromorpha* and *Ulva* for European shores than previously recognized. BLIDING's revisions are based on living samples collected from widely distant points along the European coasts, and the number of samples studied per taxon is therefore necessarely limited in relation to the vastness of these coasts.

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The question behind the present study is whether a much more intensive sampling strategy in a much more limited geographical area would either lead to the same taxonomic concepts as BLIDING's, or would produce morphological intermediates thus permitting the distinction of fewer, but geno- as well as phenotypically polymorphic species, or would disclose on the contrary, an even greater diversity on a much more local scale and this in relation to the vast estuarine gradients typical for the Netherlands coasts.

The results of a first and second study in this series of papers on the taxonomy of the Netherlands species of *Ulva* (KOEMAN & van den HOEK, 1981) and *Enteromorpha*, section *Enteromorpha* (KOEMAN & van den HOEK, 1982), respectively, largely confirm BLIDING's taxonomic concepts, but revealed an even greater diversity than found by BLIDING. Moreover, the number of species inhabiting the Netherlands coast appeared to be much greater than previously thought. The present paper reports the results of our researches on the taxonomy of *Enteromorpha*, section *Proliferae*.

In the present series on the taxonomy of *Ulva* and *Enteromorpha* in the Netherlands, we adopt as much as possible the nomenclature of BLIDING for the following reasons : BLIDING's work is the best modern treatise of the taxonomy of Ulvales, and nomenclatural changes, if necessary, should be limited to revisions for much larger areas than the Netherlands coasts.

MATERIAL AND METHODS

Material (182 living samples in the section *Proliferae* on a total of 676 samples in the genus *Enteromorpha*) was collected in the period from February 1975 through December 1977 from the stations indicated in Fig. 1, and described in Table 1 of the preceding paper on *Enteromorpha* (section *Enteromorpha*, KOEMAN & van den HOEK, 1982). For particulars of sampling and description of natural material, see KOEMAN & van den HOEK (1981).

In addition, the morphology of about 7 days old cultured germlings was studied as well as the morphology of 30 days old plants cultured in media with the following salinities : 0.5°/oo S (medium 1); 1.5°/oo S (medium 2); 4°/oo S (medium 3); 9°/oo S (medium 4); 17°/oo S (medium 5); 25°/oo S (medium 6); and 34°/oo S (medium 7). For a more complete treatment of the methods used, see KOEMAN & van den HOEK (1981); for a description of the sampling stations, and criteria used for distinction of species in *Enteromorpha*, see KOEMAN & van den HOEK (1982).

SECTION *PROLIFERAE* NOV. SECT.

Lectotype species of the genus *Enteromorpha* Link, 1820 (nom. cons.) is *E. intestinalis* (L.) Link (SILVA, 1952, p. 294; PAPENFUSS, 1962, p. 314). Type species of the section *Proliferae* is *E. prolifera* (O.F. Müller) J.G. Agardh, fig 1

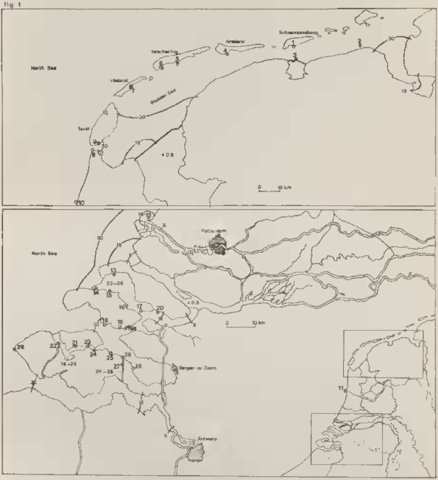


Fig. 1. – Map of the Netherlands showing stations and approximate isohalines in tidal waters and salinity ranges (°/oo S) in nontidal waters. The investigated waters were divided into the following salinity sections : 1. euhalinicum (between the 32 and 30°/oo S isohalines); 2. polyhalinicum (between the 30 and 18°/oo S isohalines). 3. mesohalinicum (between the 18 and 5°/oo S isohalines); and 4. the oligohalinicum (between the 5 and 0.5°/oo S isohalines). Moreover, some stagnant oligohaline to freshwater ditches were sampled (stations nrs : 2b, 3a, 4a, 8a, 10a, 16a, 18a, 18b, 26b).
Actually the tidal waters are subject to vast semidiurnal salinity fluctuations and fluctuations depending on river discharge, whereas the brackish man-made lakes show much less pronounced and yearly rather than dayly salinity fluctuations. Lake Grevelingen (stations 13-17) and Lake Veere (stations 21-24) used to be tidal estuaries, but were enclosed by dams and transfotmed into saline lakes in 1961 and 1971, respectively (for references see Koeman & van den Hoek, 1981).

TABLE 1

E. simplex	*E. prolifera*	*E. radiata*	*E. torta*	*E. ahlneriana*
Thalli strap-shaped, mostly unbranched, or with branches concentrated on the basal part of the thallus.	Thalli strap-shaped, branched, long strap-shaped branches often along the whole axis, small branches concentrated on the basal part of the axis.	Thalli strap-shaped to filiform, mostly densely branched and clad with numerous microscopic branchlets. Less densely branched parts often in alternation with densely branched parts.	Thalli filiform, unbranched.	Thalli strap-shaped to filiform, branched long branches often along the whole axis, small branches concentrated on the basal part of the axis.
Central cavity at least 25 µm in diameter, in small branches.	Central cavity at least 25 µm in diameter, in small branches.	Central cavity at least 25 µm in diameter, in small branches.	Central cavity 10-15 µm in diameter.	Central cavity at least 25 µm in diameter, in small branches.
3-4 celled microscopic branchlets mostly uniseriate with a big tip cell. The outer cell wall sometimes strongly thickened (Fig. 26).	3-4 celled microscopic branchlets mostly uniseriate with a big tip cell. The outer cell wall sometimes strongly thickened (Fig. 40).	3-4 celled microscopic branchlets mostly uniseriate with a big tip cell. The outer cell wall sometimes strongly thickened (Fig. 66).	Multiseriate apex without dominant tip cell, or a monoseriate apex of 2-3 cells with a relatively small tip cell.	3-6 celled microscopic branchlets mostly uniseriate, with a normal or slightly bigger tip cell. The outer cell wall not thickened.
Monoseriate apex of filiform branchlets up to 2 cells long, tip cell slightly bigger than the other cells.	Monoseriate apex of filiform branchlets up to 3 cells long, tip cell bigger than the other cells.	Monoseriate apex of filiform branchlets mostly not more than 1-2 cells long, tip cell slightly bigger than the other cells.	See above.	Monoseriate apex of filiform branchlets 1-3 cells long, sometimes longer, tip cell sometimes slightly bigger than the other cells.

Axis gradually narrowed into a long slender to rel. firm, mostly spirally twisted stipe.	Axis gradually narrowed into a long slender to rel. firm straight stipe.	Axis gradually narrowed into a fragile stipe.	In nature always without stipe.	Axis gradually narrowed into a fragile to firm stipe.
Cells in basal regions arranged in long longitudinal rows; vegetative cells in pairs. Cell walls 2-5 μm thick, thickest in the upper part of the region.	Cells in basal regions arranged in short to long longitudinal rows; veg. cells in pairs. Cell walls 2-5 μm thick, thickest in the upper part of the region.	Cells in basal regions arranged in short to long longitudinal rows; vegetative cells mostly not in pairs. Cell walls 2-3 μm thick	Cells in all regions with the same features, see below.	Cells in basal regions arranged in long longitudinal rows; vegetative cells not in pairs. Cell walls 3-6 μm thick.
Cells in middle and apical regions showing more or less equal divisions, irregularly polygonal or rectangular to quadrangular with rounded corners, arranged in rows with diverse orientation in broader parts of the thallus or in longitudinal rows in narrower parts, often 2-8 celled groups which show less order among themselves are surrounded by a thicker cell wall.	Cells in middle and apical regions showing nearly equal divisions, irregularly polygonal or rectangular to quadrangular with rounded corners, arranged in short to long longitudinal rows and short transverse rows, sometimes this order is oblique or disturbed by 2-8 celled groups bordered by thicker cell walls.	Cells in middle and apical regions showing nearly equal cell divisions, polygonal, rectangular or quadrangular, often round or rounded with 4-6 corners; arranged in longitudinal rows or in pairs which are arranged in longitudinal rows, sometimes with an indication of transverse rows, or unordered.	Cells showing equal cell divisions, mostly rectangular or quadrangular, arranged in longitudinal rows, sometimes an oblique orientation in relation to the direction of the thread.	Cells in middle and apical regions showing equal divisions, rectangular or quadrangular, arranged in longitudinal and transverse rows.

1883, p. 129. The section *Proliferae* largely agrees with BLIDING's (1963, p. 45) «Prolifera Group», however, *E. torta* (Mertens) Reinbold is here included in this section. Bliding places *E. torta* in the «Torta Group».

Cells in middle region varying from about 10 x 7 µm to 17 x 13 µm. Cells in apical and middle region showing mostly equal divisions, and being arranged in longitudinal and often transverse cell rows or in 4-8 celled groups. The central thicker part of the parietal chloroplast containing the mostly single pyrenoid usually situated centrally against the peripheral cell wall. Thallus filiform to linear, the broader thalli oblong in outline, mostly compressed, the two layers loosely adnate, leaving a hollow margin, or tubular; unbranched or with main branches (having the form of the axis) and branchlets concentrated towards the basal region, or along the whole axis.

Latin diagnosis :

Cellulae regionis medianae magnitudine 10 x 7 usque ad 17 x 13 µm. Regionis apicalis et medianae cellulae divisionibus plerumque aequalibus in seriebus longitudinalibus et saepe transversalibus sive in gregibus 4-8 cellularum dispositae. Chloroplastus integumentum cellulare externum tegens una plerumque pyrenoide. Thallus filiformis – linearis – oblongus, plerumque complanatus, cellularum duobus stratis laxe adnatis marginibus cavis; sive tubulosus; simplex vel ramis forma axium et ramulis praecipue in axis regione basali seu extendentibus secus axes totas.

Identification of species in the section *Proliferae*

Identification of the five species of *Enteromorpha* in the section *Proliferae* is facilitated by a table which permits the comparison of a combination of characters (Table 1). As in the section *Enteromorpha*, the macroscopic morphology of plants presents the most distinctive differences between the species of the section *Proliferae*. In *E. ahlneriana*, the mode of reproduction is characteristic, and to a lesser degree the morphology of filiform branchlets. The remaining microscopic characters are also less distinctive and are subject to wide variation and overlap. *E. simplex* is mostly characterized by the spirally twisted stipe.

— ***E. SIMPLEX*** **(Vinogradova) nov. comb. nov. stat.**

E. prolifera f. *simplex* Vinogradova, 1974, p 99; *E. prolifera* (O.F. Müller) J.G. Agardh subsp. *prolifera* typus I Bliding, 1963, p. 46.

- Description (Figs 2-33)

Morphology (Figs 2-12)

Thalli oblong, strapshaped, or filiform, the two layers compressed or loosely adnate with hollow margins, more or less wrinkled and lubricous, light to medium green, unbranched or less commonly with primary branches concentrated in the basal part of the frond. Most branches short in relation to the main axis, branches filiform to linear. Apex of main axis, obtuse, often opened. Basis attenuate, stipe long, slender to relatively firm, mostly spirally twisted, with a small disciform holdfast. Margins entire, in the upper part often undulate. Length up to 15 cm and 1.5 cm broad, branches mostly not longer than 3 cm In general plantes are not longer than 8 cm and up to 5 mm broad.

Anatomy, lower basal region (Figs 11, 28, 29)

In surface view, the pale coloured relatively long and mostly spirally twisted stipe shows rounded or elongate rhizoidal cells with rounded corners in the lowest region which have the same size and colour as normal vegetative cells in this region, or are only slightly larger and with darker contents. In this part cells are mostly arranged in short or long longitudinal rows, also vegetative cells may be united in pairs. Cell walls 2-5 μm thick, exceptionally thicker, thickest in the upper part of this region. Chloroplast parietal, often any structure obscured by numerous large starch grains. Bases of the younger branches without, of the older and bigger ones with rhizoidal cells, which are often long and clearly visible (Fig. 33).

Anatomy, upper basal region (Fig. 30)

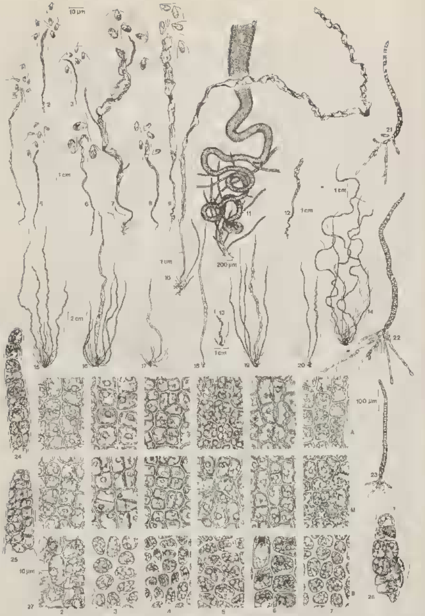
Cells in the upper basal region irregularly polygonal, with 3-5 rounded corners, showing mostly more or less equal divisions, arranged in often distinct longitudinal and sometimes transverse rows, or in pairs which may form short undulating rows. Cell walls 2-4 μm thick. The central thicker part of the parietal chloroplast, containing the pyrenoid(s) sometimes slightly tilted towards any anticlinal cell wall, but mostly situated centrally against the peripheral cell wall. Chloroplast arms relatively thick, descending along the anticlinal cell walls. The chloroplast structure in this part is often obscured by numerous large starch grains. Pyrenoids one per cell, two in less than 10% of the cells, 3-5 μm in diameter, round.

Anatomy, middle and apical region (Figs 31, 32)

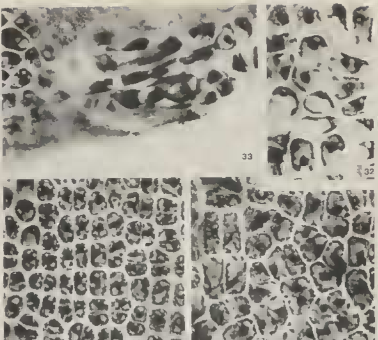
Cells in the middle and apical region irregularly polygonal, to regularly rectangular or quadrangular, with rounded corners, showing more or less equal divisions and arranged in rows with diverse orientation in broader parts of the lamina or in longitudinal rows in the narrower parts; often the cells are arranged in 2-8 celled groups, which show less order among themselves, and are recognized by their common thick cell wall. Cell walls 0.5-2 μm thick. The central thicker part of the parietal chloroplast, containing the pyrenoid(s), sometimes slightly tilted towards any anticlinal cell wall, or in the apical region with a preference for the apically oriented one, but mostly situated centrally against the peripheral cell wall. Descending arms often thick, descending along the anticlinal cell walls. The chloroplast structure is often obscured by numerous large starch grains. Pyrenoids one par cell, two in less than 10% of the cells, 2-4 μm in diameter, round.

Anatomy, tips of short filiform branches (Figs 24-26)

In surface view, the monoseriate apex of young branches is very short, mostly only two cells long. The tip-cell is slightly bigger than the other cells, which are on the whole somewhat smaller compared to normal thallus cells.



Figs 2-27. – *Enteromorpha simplex.* Figs 2, 3, 4, 5. male gametophytes with gametes, plants no. 175, 176, 303, 167. Figs 6, 7. sporophytes with zoospores, plants no. 183, 179. Figs 8, 9. female gametophytes with gametes, plants no. 253, 173. Fig. 10. plant no. 543. Fig. 11. basis of plant no. 178. Fig. 12. plant no. 66. Figs 13, 14. 30 days old cultures, same material as Fig. 12. Fig. 13 in 17‰ S medium, Fig. 14 in 34‰ S medium. Figs 15, 16, 17, 18, 19, 20. 30 days old cultures, same material as Fig. 10. Fig. 15 in 1.5‰ S medium, Fig. 16 in 4‰ S medium, Fig. 17 in 9‰ S medium, Fig. 18 in 17‰ S medium, Fig. 19 in 25‰ S medium, Fig. 20 in 34‰ S medium. Figs 21, 22, 23. 7 days old germlings. Figs 24, 25, 26. branchlets, Fig. 24 with monoseriate apical cell row, Figs 25, 26 with an apical cell only, Fig. 26 with strongly thickened cell wall. Fig. 27. Cells in surface view, same material as Figs 15-20; B = basal region, M = middle region, A = apical region. 2, 3, 4, 5, 6, 7, refer to media (see material and methods).



Figs 28-33. – *E. simplex*, cells in surface view, same material as Fig. 10. Fig. 28. lower basal region, lower zone. Fig. 29. lower basal region, upper zone. Fig. 30. upper basal region. Fig. 31. middle region. Fig. 32. apical region. Fig. 33. basis of branch with rhizoidal cells.

Very young branchlets of 3-4 cells mostly uniseriate, and with a bigger tip-cell. The outer cell wall is often strongly thickened.

Reproductive cells (Figs 2-9)

In nature most plants are dioecious gametophytes, producting 2-flagellate gametes, the male ones slightly smaller and with smaller chloroplasts than the female ones. They are positively phototactic, and are able to germinate in small amounts without fertilization. Sporophytes produce big 4-flagellate zoospores, which are also positively phototactic.

Measurements, based on 66 plants :

axis, cells in surface view :	
lower basal region	(14-)18(-22) x (10-)11(-13) μm
lower basal region, upper zone	(11-)13(-16) x (8-)9(-11) »
upper basal region	(10-)12(-14) x (7-)9(-10) »
middle region	(10-)12(-14) x (8-)9(-11) »
apical region	(11-)14(-16) x (8-)10(-11) »
– branches, cells in surface view :	
basal region	(10-)12(-14) x (7-)9(-10) »
middle region	(9-)11(-13) x (7-)8(-10) »
apical region	(9)11(-13) x (6-)8(-10) »
male gametes	(5-)6(-7) x (3.5-)4(-4.5) »
female gametes	(7-)8(-9) x (5-)5.5(-6) »
– zoospores	(8-)9.5(-11) x (4-)5.5(-6.5) »

Morphology of germlings and young fronds (Figs 21-23)

Zoospores and gametes germinate by forming a rhizoid, immediately after that a strongly growing upright monoseriate filament. Sometimes the rhizoidal part remains small and contains densely branched rhizoids. From some special rhizoidal cells often more filaments are formed. In a later stage these filaments grow into hollow cylinders, and the stipe may spirally wind, especially at the higher salinities tested.

- Ecology and distribution

E. simplex has been collected from 22 stations (see Table 2, and Fig. 1).

TABLE 2. Specimens investigated
(for locality numbers see Table 1, list of localities, in KOEMAN & van den HOEK, 1982)

1 (II, '75, plants no. 41, 42; V, '75, plants no. 17, 80, 81) 1a (VII, '75, plant no. 213); 2 (VIII, '75, plant no. 66); 3 (V, '76, plant no. 623); 5 (III, '76, plants no. 239, 252, 253; V, '76, plants no. 469, 481, 482, 484); 5a (III, '76, plant no. 259), 6a (III, '76, plant no. 263); 6b (VI, '75, plants no. 162, 165, 166, 167; VI, '76, plants no. 429, 430); 6c (VI, '75, plants no. 173, 174, 175, 176, 177, 178, 179, 180, 181, 182, 183). 7 (III, '76, plants no. 214, 215; VI, '76, plant no. 502); 7a (III, '76, plant no. 227a); 7c (III, '76, plant no. 231); 9 (V, '76, plant no. 636); 16 (IV, '76, plants no. 421, 422); 19 (IV, '76, plant no. 420a); 19a (VI, '75, plant no. 205); 22 (IV, '76, plants no. 333, 334, 335); 22a (IV, '76, plant no. 322); 23 (IV, '76, plants no. 303, 306); 26 (VI, '75, plant no. 114, IV, '76, plants no. 355, 356, 358); 27 (IV, '76, plants no. 425, 426); 28 (IV, '76, plants no. 363, 364, 369, 371, 372, 373, 381, 383).

Ten of these were littoral to high littoral mud or sandy mudflats, where the species was found growing attached to all kinds of solid substrates, often in dense populations, the thin spirally twisted bases entangled between each other. Four localities were sheltered littoral and upper sublittoral zones on seadikes, harbour moles and in oysterponds, where the species was growing on stones and shells as well as epiphytically on *Fucus vesiculosus*. Two localities were a breakwater and a harbour mole exposed to strong wave action, where the species was growing on the same substrates in the same littoral zones. Finally the species was encountered in six localities in meso- or polyhaline semi-stagnant or stagnant saline lakes or canals. This accords with the results of culture experiments, which gave good growth of germlings and young fronds in media with salinities ranging from 34-9o/oo S (plant no. 543 still grew well at 1.5o/oo S, Figs 13-20).

- Morphological and anatomical characters in cultures (Figs 13-20, 27)

The important morphological and anatomical characters were retained in cultures; however, cultured plants were more branched, even in the offspring of an unbranched plant, and the thalli were narrower than in the original material. The characteristic spirally twisted stipe was well developed, especially at higher salinities. Most cells in the middle and apical region contained one central pyrenoid per cell, the parietal chloroplast completely covering the outer cell wall, in the basal parts sometimes slightly tilted towards any anticlinal cell wall. Cells were on the whole well ordered especially in the apical region of the thallus, and groups of 4-8 cells often shared a common thicker cell wall, like in the wild material. Plants cultured in low salinity media had about the same size of cells as those in high salinity media (Fig. 27, and Table 3).

TABLE 3

wild material of *E. simplex* 541	medium	cultures
upper basal region (11-)14(-16) x (7-)10(-12)	7	(9-)11(-13) x (6-)8(-10)
	5	(8-)10(-13) x (5-)7(-9)
	4	(14-)17(-20) x (11-)12(-14)
	3	(14-)16(-17) x (9-)11(-14)
	2	(12-)15(-17) x (8-)11(-13)
middle region (9-)10(-11) x (7-)8(-9)	7	(8-)10(-13) x (8-)9(-9)
	5	(6-)8(-10) x (7-)8(-9)
	4	(10-)13(-15) x (6-)9(-11)
	3	(12-)14(-16) x (9-)11(-13)
	2	(10-)12(-13) x (7-)9(-11)
apical region (10-)12(-13) x (9-)10(-11)	7	(7-)9(-11) x (6-)7(-8)
	5	(9-)11(-13) x (7-)9(-10)
	4	(9-)10(-11) x (6-)8(-10)
	3	(11-)13(-16) x (8-)10(-13)
	2	(8-)12(-15) x (6-)8(-9)

TABLE 4

wild material of *E. simplex* 66	medium	cultures
upper basal region (9-)10(-12) x (7-)8(-9)	7	(12-)14(-17) x (8-)10(-11)
middle region (10-)11(-13) x (7-)9(-10)	7	(9)11(-13) x (7-)9(-11)
apical region (9-)12(-12) x (7-)8(-10)	7	(9-)12(-14) x (7-)8(-10)

TABLE 5

wild material of *E. simplex* 543	medium	cultures
upper basal region (10-)12(-13) x (7-)9(-10)	7	(8-)10(-12) x (7-)8(-9)
	6	(13-)17(-21) x (9-)11(-14)
	5	
	4	(9-)10(-11) x (6-)8(-9)
	3	(10-)12(-13) x (8-)10(-11)
	2	(15-)18(-21) x (-1)14(-17)
middle region (11-)13(-14) x (8-)10(-11)	7	(11-)13(-15) x (8-)10(-11)
	6	(11-)13(-16) x (8-)10(-11)
	5	(14-)16(-19) x (9)11(-13)
	4	(10)12(-14) x (7-)9(-11)
	3	(9-)12(-16) x (8-)10(-13)
	2	(10-)12(-15) x (9-)11(-12)
apical region (15-)17(-20) x (10-)12(-14)	7	(10-)12(-15) x (9-)10(-12)
	6	(9-)12(16) x (7-)10(-12)
	5	(14-)16(-18) x (10-)13(-15)
	4	(9-)12(-14) x (7-)10(-12)
	3	(14-)16(-18) x (12-)14(-16)
	2	(12-)14(-16) x (9-)11(-13)

Tables 3, 4, 5. — Cell sizes in μm of *E. simplex*, plants no. 541, 66, and 543 (wild material) and of 30 days old cultures isolated from these plants and kept at different salinities (7 = 34‰ S; 6 = 25‰ S; 5 = 17‰ S; 4 = 9‰ S, 3 = 4‰ S 2 = 1.5‰ S).

— ***E. PROLIFERA*** **(O.F. Müll.) J. Ag.**

E. prolifera (O.F. Müller) J.G. Agardh 1883, p. 129; *E. prolifera* (O.F. Müller) J.G. Agardh subsp. *prolifera* typus II Bliding 1963, p. 50.

- Description (Figs 34-53)

Morphology (Figs 34-38, 41)

Thalli strapshaped, the two layers compressed or loosely adnate, mostly with hollow undulating margins, more or less wrinkled and lubricous, light to yellowish green on high littoral marine sand- and mudflats to medium green in more shaded places. Plants with branches of the first order, small branches concentrated in the basal region, long strapshaped branches often higher up along the axis, sometimes longer than the main axis. Apices of main axis and long branches obtuse, often open. Basis attenuate, stipe long, slender to relatively firm, with a small disciform holdfast, from which diminutive new thalli may arise. Plants up to 50 cm long, axis up to 30 cm long and 10 mm broad, branches up to 30 cm long and 5 mm broad.

Anatomy, lower basal region (Figs 38, 49, 50)

In surface view, the relatively long and slender stipe shows big dark coloured and often elongate rhizoidal cells in the lowest region which have the same morphology and colour as normal vegetative cells in this region. In this part cells are mostly arranged in short or long longitudinal rows; vegetative cells may be united in pairs. Cell walls 2-5 µm thick, thickest in the upper part of this region. Chloroplast parietal, mostly any structure obscured by numerous large starch grains. Bases of the younger branches without, of the older and bigger ones with rhizoidal cells.

Anatomy, upper basal region (Fig 51)

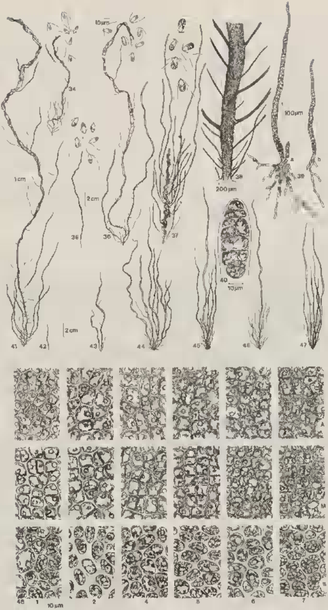
Cells in the upper basal region irregularly polygonal, with 4-6 rounded corners, showing mostly more or less equal divisions, arranged in often distinct longitudinal and sometimes short transverse rows, or in pairs which may form short longitudinal rows. Cell walls 2-4 µm thick. The central thicker part of the chloroplast, containing the pyrenoid(s) sometimes slightly tilted towards any anticlinal cell wall, but generally situated centrally against the peripheral cell wall. Descending arms relatively thick, descending along the anticlinal cell walls. The chloroplast structure in this part is often obscured by numerous large starch grains. Pyrenoids one per cell, two in less than 10% of the cells, 3-5 µm in diameter, round.

Anatomy, middle and apical region (Figs 52, 53)

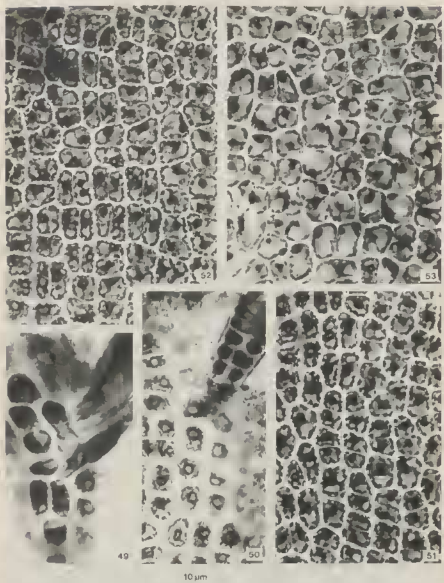
Cells in the middle and apical region irregularly polygonal, or rectangular to quadrangular in some parts of these regions, with more or less rounded corners, showing nearly equal divisions and arranged in short to long longitudinal rows, and shorter transverse rows; sometimes this ordering is oblique or is disturbed by small fields consisting of less ordered cells. 2-8 celled groups are often bordered by thicker cell walls. Cell walls 0.5-2 µm thick. The central thicker part of the chloroplast, containing the pyrenoid(s) sometimes slightly tilted towards any anticlinal cell wall, but generally situated centrally against the peripheral cell wall, sometimes with an additional lobe connecting the lateral arms. Descending arms often thick, descending along the anticlinal cell walls. The chloroplast structure is often obscured by numerous large starch grains. Pyrenoids one per cell, two in less than 10% of the cells, 2-4 µm in diameter, round.

Anatomy, tips of short filiform branches (Fig. 40)

In surface view, the monoseriate apex of young branches is short, mostly not more than three cells long. The tip cell is slightly bigger than the other



Figs 34-47. – *E. prolifera*. Fig. 34. male gametophyte with gametes, plant no. 102. Figs 35, 36. female gametophytes with gametes, plants no. 112, 204. Fig. 37. sporophyte with zoospores, plant no. 227. Fig. 38. basis of plant no. 102. Fig. 39 a, b. 7 days old germlings, same material as Fig. 41. Fig. 40. branchlet with short monoseriate apical cell row. Fig. 41. plant no. 542. Figs 42, 43, 44, 45, 46, 47. 30 days old cultures, same material as Fig. 41. Fig. 42 in 0.5°/oo S medium, Fig. 43 in 1.5°/oo S medium, Fig. 44



in 9°/oo S medium, Fig. 45 in 17°/oo S medium, Fig. 46 in 25°/oo S medium, Fig. 47 in 34°/oo S medium. Fig. 48. cells in surface view, same material as Figs 42-47, B = basal region, M = middle region, A = apical region. 1, 2, 4, 5, 6, 7, refer to media (see material and methods).

Figs 49 53. – *E. prolifera*, cells in surface view, same material as Fig. 41. Fig. 49. lower basal region, lower zone. Fig. 50. lower basal region, upper zone. Fig 51. upper basal region. Fig. 52. middle region. Fig. 53. apical region.

cells. 3-4 celled microscopic branchlets are mostly uniseriate and have a bigger tip cell. The outer cell wall is often strongly thickened basally.

Reproductive cells (Figs 34-37)

In nature most plants are dioecious gametophytes, producing 2-flagellate gametes, the male ones slightly smaller and with smaller chloroplasts than the female ones. They are positively phototactic, but may become negatively phototactic after some time, and are able to germinate without fertilization. Sporophytes produce big 4-flagellate zoospores which are positively phototactic

Measurements based on 33 plants

– axis, cells in surface view :		
lower basal region	(11-)13(-16) x (8-)10(-11)	μm
lower basal region, upper zone	(10-)12(-14) x (7-)9(-10)	»
upper basal region	(10-)12(-14) x (7-)9(-10)	»
middle region	(10-)12(-13) x (7-)8(-10)	»
apical region	(11-)14(-16) x (8-)10(-12)	»
– branches. cells in surface view :		
basal region	(11-)14(-16) x (9-)10(-12)	»
middle region	(9-)12(-14) x (7-)9(-10)	»
apical region	(12-)14(-15) x (9-)11(-12)	»
– male gametes	(4-)5(-6) x (3-)4(-4.5)	»
– female gametes	(5-)5.5(-6.5) x (3.5-)4(-4.5)	»
zoospores	(8-)9(-10.5) x (4-)5(-6.5)	»

Morphology of germlings and young fronds (Fig 39)

Zoospores and gametes germinate by forming a rhizoid, immediately after that a strongly growing upright monoseriate filament. Sometimes, as in the preceding species, the rhizoidal part remains small and consists of densely branched rhizoids. From some spherical rhizoidal cells often more filaments are formed. In a later stage the filaments grow into hollow cylinders. The first formed frond, the main axis may form, especially at higher salinities, branches on the basal part.

- Ecology and distribution

E. prolifera has been collected from 13 stations (see Table 6, and Fig. 1).

TABLE 6. – Specimens investigated
(for locality numbers see Table 1, list of localities, in KOEMAN & van den HOEK, 1982)

1 (II, '75, plants no. 36, 37, 38, V, '75, plants no. 21, 22, 25); 2(V, '75, plant no. 28); 3a (IV, '75, plants no. 1, 2); 5 (III, '76, plant no. 240; VI, '76, plant no. 542); 6 (III, '76, plant no. 282); 7 (III, '76, plants no. 216, 217, 218, 219; V, '76, plant no. 446; VI, '76, plants no. 499, 500); 7a (III, '76, plants no. 227, 228a; VI, '76, plants no. 504); 8a (VI, '75, plant no. 76); 19a (VI, '75, plants no. 204, 207); 23 (IV, '76, plant no. 304); 26 (VI, '75, plant no. 112); 26b (VI, '75, plants no. 73a, 74a, 74c); 27 (VI, '75, plants no. 101, 102, 103).

Six of these were littoral to high littoral mud or sandy mud flats where the species was found growing attached to stones or shells, two localities were harbour moles exposed to strong wave action, where the species was growing epiphytically on *Fucus vesiculosus* as well as on stones and shells. Two other localities were a meso- or polyhaline semi-stagnant or stagnant saline lake and canal; finally the species was encountered in three localities growing attached to phanerogams and wooden poles, and free floating in oligo- to mesohaline canals and pools: often the plants were, in such places, inflated and light green when full-grown, resembling *E. intestinalis* or *E. pilifera* except in the way of branching. This accords with the results of culture experiments, which gave good to very good growth of germlings and young fronds in media with salinities ranging from 34-1.5‰ S (Figs 42-47).

- Morphological and anatomical characters in cultures (Figs 42-48)

The important morphological and anatomical characters were retained in cultures; however the thalli were on the whole narrower, and branching was less dense and more concentrated on the basal part of the axis, especially at higher salinities, than in the original material. Spirally twisted stipes did not occur. Most cells in the middle and apical region contained one central pyrenoid per cell. The parietal chloroplast was completely covering the outer cell wall. Cells in middle and apical regions tend to be slightly smaller than in the wild material and were well ordered in these regions of the thallus. Groups of 4-8 cells often shared a common thicker cell wall, like in the wild material (Fig. 48, Tables 7, 8).

TABLE 7

wild material of *E. prolifera* 542	medium	in culture, cell dimensions
upper basal region (13-)16(-19) x (7-)10(-12)	7	(9-)11(-13) x (8-)9(-10)
	6	(9-)12(-14) x (7-)9(-11)
	5	(11-)13(-15) x (7-)9(-10)
	4	(11-)14(-19) x (8-)10(-11)
	3	(13-)16(-20) x (7-)10(-12)
	2	(10-)12(-14) x (7-)9(-10)
	1	(9-)11(-12) x (7-)8(-9)
middle region (13-)16(-18) x (8-)11(-13)	7	(7-)9(-11) x (5-)7(-8)
	6	(7-)9(-10) x (5-)7(-8)
	5	(8-)10(-11) x (-6)7(-8)
	4	(8-)11(-13) x (6-)8(-9)
	3	(8-)10(-12) x (5-)7(-8)
	2	(9-)11(-13) x (7-)9(-10)
	1	(8-)10(-11) x (6-)7(-8)
apical region (13-)15(-17) x (11-)13(-15)	7	(8-)10(-12) x (6-)8(-9)
	6	(8-)10(-11) x (6-)8(-9)
	5	(7-)10(-13) x (7-)8(-9)
	4	(11-)13(-15) x (7-)9(-11)
	3	(9-)12(-12) x (8-)9(-10)
	2	(10-)12(-14) x (7-)9(-10)

TABLE 8

wild material of *E. prolifera* 111	medium	in culture, cell dimensions
upper basal region (11-)13(-14) x (7-)8(-10)	7 6 5	(12-)14(-16) x (9-)10(-10) (13-)15(-17) x (7-)10(-13) (15-)20(-25) x (9-)12(-15)
middle region (9-)11(-12) x (7-)8(-9)	7 6 5	(10-)11(-12) x (6-)7(-8) (12-)13(-15) x (9-)11(-12)
apical region (9-)11(-13) x (7-)8(-9)	7 6 5	(9-)10(-12) x (6-)8(-9) (9-)10(-12) x (6-)8(-9) (14-)17(-20) x (12-)13(-14)

Tables 7, 8. — Cell sizes in μm of *E. prolifera*, plants no. 542 and 111 (wild material) and of 30 days old cultures isolated from these plants and kept at different salinities (7 = 34‰ S; 6 = 25‰ S; 5 = 17‰ S; 4 = 9‰ S; 3 = 4‰ S; 2 = 1.5‰ S; 1 = 0.5‰ S).

— *E. RADIATA* **J. Ag.**

E. radiata J.G. Agardh 1883, p. 156; *E. prolifera* subsp. *radiata* (J.G. Agardh) Bliding 1963, p. 56.

- Description (Figs 54-78)

Morphology (Figs 54-58, 67)

Thalli strapshaped, linear to filiform, in broader parts of the thallus the two layers compressed or loosely adnate with hollow margins, mostly strongly wrinkled; narrower parts of the thallus completely hollow. Plants medium to dark green, densely branched. Every branch of sufficient length branched again or remaining nearly unbranched. Branches and main axis with the same morphology. The apical parts of very long branches and main axis often broad and less branched than more basal parts, which are often densely clad with very small branches and branchlets; however, along axis and long branches these different zones may occur in alternation. The axis gradually narrowed, towards the base, into a fragile stipe with a small disciform holdfast; but mostly the basal parts are buried in the sediment and originate by vegetative propagation from old buried thalli. Apices of axis and long branches truncate to acute, often open or damaged. Plants up to 2 m long, up to 1 cm broad, but mostly not more than 2-3 mm broad.

Anatomy, lower basal region (Figs 67, 73, 75, 78)

In surface view, the long relatively firm or slender stipe, which has the same morphology as the bases of long branches, shows dark coloured rounded rhizoidal cells, which are more elongate towards the base, in the lowest region, and which have the same morphology and colour as normal vegetative cells in this region, or are somewhat darker. In this part cells are mostly arranged in short or long longitudinal rows. Cell walls 2-3 µm thick. Chloroplast parietal, mostly any structure obscured by numerous large starch grains; sometimes, in small fields of cells, the chloroplasts are tilted towards the apically oriented cell wall. Bases of all branches with rhizoidal cells (Fig. 78), except very small branchlets. The central cavity often strengthened by transverse trabeculae (Fig. 75).

Anatomy, upper basal region (Fig. 74)

Cells in the upper basal region irregularly polygonal or rectangular to rounded, with 4-6 corners, showing equal divisions, arranged in mostly distinct longitudinal rows. Cell walls 2-4 µm thick. The central thicker part of the chloroplast containing the pyrenoid(s) sometimes slightly to strongly tilted towards the apically oriented cell wall, giving the chloroplast a cap-like appearance, but generally situated centrally against the peripheral cell wall. Descending arms thin or thick, descending along the anticlinal cell walls. The chloroplast structure is often obscured by numerous large starch grains. Pyrenoid(s) one per cell, two in less than 10% of the cells, 3-5 µm in diameter, round.

Anatomy, middle and apical region (Figs 76, 77)

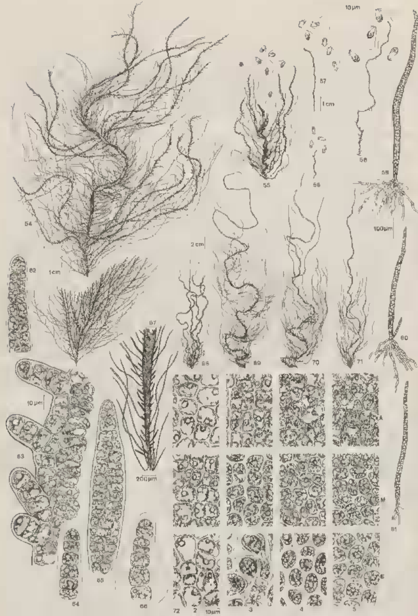
Cells in the middle and apical region irregularly polygonal or rectangular to quadrangular in small fields, often round or rounded with 4-6 corners, showing nearly equal divisions, and arranged in mostly distinct longitudinal rows, or in pairs, which are arranged in longitudinal series; in broader parts often with an indication of transverse rows, however in some parts or plants cells are predominantly unordered. Cell walls 0.5-2 µm thick. The central thicker part of the chloroplast, containing the pyrenoid(s) sometimes slightly tilted towards any anticlinal cell wall or predominantly towards the apically oriented one, but generally situated centrally against the peripheral cell wall. Descending arms thin or thick, descending along the anticlinal cell walls. The chloroplast structure is often obscured by numerous large starch grains. Pyrenoid(s) one per cell, two in less than 10% of the cells, 3-5 µm in diameter.

Anatomy, tips of short filiform branches (Figs 62-66)

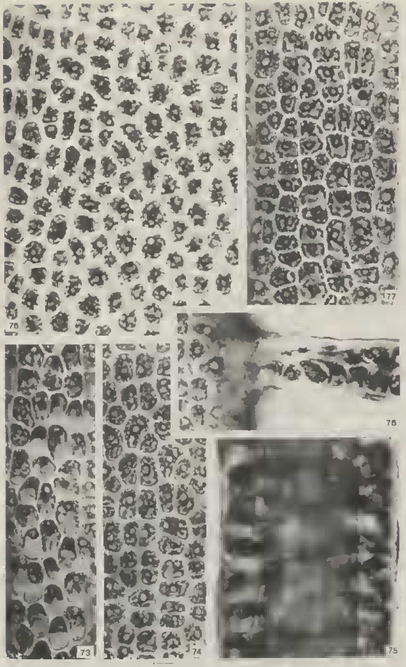
In surface view, the monoseriate apex of young branches is very short, mostly consisting of one relatively big tip-cell, and of cells below it which are often short and broad. 3-4 celled microscopic branchlets are mostly uniseriate and have bigger tip-cells. The outer cell wall is sometimes strongly thickened basally.

Reproductive cells (Figs 55-58)

In nature, only very small parts of the apices become fertile, but often it was impossible to obtain reproductive cells from some plants. Plants which grew fertile were gametophytes, producing 2-flagellate gametes, the male ones slightly smaller and with smaller chloroplasts than the female ones, or sporophytes, producing big 4-flagellate zoospores. Gametes and spores were positively phototactic, and gametes were able to germinate in small amounts.



Figs 54-72. *E. radiata*. Fig. 54, plant no. 531. Figs 55-56. male gametophytes with gametes, plants no. 444, 224. Fig. 57. female gametophyte, with gametes, plant no. 230. Fig. 58. sporophyte with zoospores, plant no. 506. Figs 59, 60, 61. 7 days old germlings. Figs 62, 63, 64, 65, 66. young branchlets, Fig. 62 with an apical cell only, Figs 63, 64, 65, 66 with monoseriate apical cell row, Fig. 66 with strongly thickened cell wall. Fig. 67. basis of plant no. 168. Figs 68, 69, 70, 71 30 days old cultures, same material as Fig. 54, Fig. 68 in 1.5‰ S medium, Fig. 69 in 4‰ S medium, Fig. 70 in 17‰ S medium, Fig. 71 in 34‰ S medium. Fig. 72. cells in surface view, same material as Figs 68-71, B = basal region, M = middle region, A = apical region. 2, 3, 4, 5, refer to media (see material and methods).



Figs 73-78. – *E. radiata*, cells in surface view, same material as Fig. 54. Fig. 73. lower basal region. Fig. 74. upper basal region. Fig. 75. lower basal region, central cavity with trabeculae. Fig. 76. middle region. Fig. 77. apical region. Fig. 78. basis of branchlet with rhizoidal cells.

Measurements based on 54 plants

– axis and long branches, cells in surface view :	
lower basal region	(13-)15(-18) x (9-)10(-12) μm
upper basal region, lower zone	(12-)14(-16) x (8-)10(-12) »
upper basal region, upper zone	(11-)13(-15) x (8-)9(-11) »
middle region	(11-)13(-16) x (8-)10(-11) »
apical region	(11-)13(-16) x (8-)10(-11) »
male gametes	(5.5-)6(-6.5) x (3.5-)4(-4.5) »
– female gametes	(6-)6.5(-7) x (3.5-)4.5(-5) »
– zoospores	(8.5-)10(-11) x (5-)6(-6.5) »

Morphology of germlings and young fronds (Figs 59-61)

Zoospores and gametes germinate by forming a small rhizoid, immediately after that a strongly growing upright monoseriate filament, which very soon becomes a hollow cylinder by longitudinal divisions. The rhizoidal part mostly remains small. From some rhizoidal cells often more filaments were formed. The first formed frond, the main axis, soon forms branches on the basal part.

- Ecology and distribution

E. radiata has been collected from 15 stations (see Table 9, and Fig. 1).

TABLE 9. Specimens collected
(for locality numbers see Table 1, list of localities, in KOEMAN & van den HOEK, 1982)

1 (II, '75, plants no. 39, 40; V, '75, plant no. 23); 2 (V, '75, plants no. 29, 31, 118; VIII, '75, plant no. 65; XII, '76, plants no. 613, 614); 2a (VI, '75, plants no. 209, 210); 5 (V, '76, plants no. 478, 485; XI, '76, plants no. 606, 607, 608); 5a (III, '76, plants no. 257, 258; V, '76, plants no. 486, 487); 6a (III, '76, plants no. 260, 261, 262, 268, 269); 6b (VI, '75, plants no. 168, 169, 170, 171, 172; VI, '76, plants no. 531, 535); 6c (VI, '75, plants no. 184, 185, 186); 7 (III, '76, plants no. 223, 224; V, '76, plants no. 443, 444); 7a (III, '76, plants no. 228, 229, 229a, 230. V, '76, plant no. 457); 9 (V, '76, plant no. 634), 18 (VI, '75, plants no. 134, 135); 19a (VI, '75, plants no. 202, 203, 208); 20 (VI, '75, plants no. 638, 639); 28 (VI, '75, plants no. 132, 133).

Thirteen of these stations were littoral to high littoral mud or sandy mudflats, where the species was found growing partly embedded in the sediment, interwoven between the basal parts of *Spartina* spec., *Salicornia* spec. and other phanerogams. These plants had often a less branched and more filiform morphology, and they were seldom attached to shells or other solid substrates. Such plants can be easely confused with *E. torta*. Often, large plants are transported by wave action and tidal currents to other places, especially floodmarks and gully banks, sometimes in big quantities. They are covered by autumn and winter storms with sediment, which the species can survive, even under nearly anaerobic conditions. In spring it proliferates with typical short and relatively broad, nearly unbranched thalli. This is later followed by strong growth of filiform thalli, which have the typical *E. radiata* morphology at a later stage. Two localities were pools on the seaside of the dike, where the species was growing in the sublittoral zone, attached to stones. It was never found growing in meso- or polyhaline semistagnant or stagnant saline lakes or canals; however, culture experiments indicate a potentially more euryhaline distribution. Germlings and young fronds grew very well in media with salinities ranging from 9-34‰ S, and grew poorly to reasonably at salinities ranging from 1.5-9‰ S (Figs 68-71).

Morphological and anatomical characters in cultures (Figs 68-72)

The important morphological and anatomical characters were retained in cultures. The different morphological types grown in cultures at different salinities reflected the broad morphological variability met in nature. However, it was for practival reasons not possible to grow plants with the dimensions of the original material. Plants cultured in media with salinities ranging from 25-34‰ S morphologically agreed best with the parent plants. In media with salinities ranging from 17‰-1.5‰ S branching was less dense, and branches were mostly not clearly concentrated in a more basal zone than to other zones. At a salinity of 4‰ S long unbranched filiform branches were abundantly produced. At 1.5‰ S two cultures of *E. radiata* (from plants no. 599 and 531) behaved differently. The first one produced tiny threads, which showed many microscopic branchlets, possibly originating from akinetes, the second one produced relatively broad basally branched thalli. Most cells in middle and apical regions contained one central pyrenoid per cell, and the parietal chloroplast mostly covered the outer cell wall, and was sometimes tilted towards an anticlinal cell wall, or predominantly to the apically oriented one. Most cells in middle and apical regions of the main axis and broader branches formed short to long longitudinal rows; in densely branched parts often no order was observed. Plants cultured in low salinity media had larger cells than those in high salinity media, which were somewhat smaller than in the wild material (Fig. 72, Tables 10 and 11).

TABLE 10

wild material of *E. radiata* 599	medium	cultures
upper basal region (10-)13(-16) x (9-)10(-11)	7	(12-)14(-16) x (10-)12(-13)
	6	(13-)15(-17) x (10-)12(14)
	5	(11-)14(-17) x (8-)10(-12)
middle region (12-)14(-16) x (10-)11(-12)	7	(9-)13(-16) x (8-)10(-11)
	6	(9-)12(-14) x (8-)9(-9)
	5	(10-)12(-14) x (8-)10(-11)
	4	(13-)15(-18) x (11-)13(-15)
	2	(13-)15(-17) x (8-)10(-12)
apical region (12-)14(-17) x (9-)11(-12)	7	(11-)13(-15) x (7-)9(-11)
	6	(9-)12(-14) x (7-)8(-9)
	5	(10-)12(-14) x (8-)9(-11)
	4	(12-)14(-16) x (7-)9(-11)

TABLE 11

wild material of *E. radiata* 531	medium	cultures
upper basal region (13-)17(-20) x (9-)10(-12)	7	(9-)11(-12) x (8-)9(-10)
	5	(10-)13(-15) x (9-)11(-12)
	3	(11-)19(-28) x (8-)12(-14)
	2	(12-)16(-21) x (10-)14(-16)
middle region (11-)15(-18) x (9-)11(-12)	7	(9-)11(-13) x (7-)9(-10)
	5	(9-)11(-13) x (7-)9(-10)
	3	(11-)12(-14) x (9-)10(-11)
	2	(10-)12(-14) x (8-)10(-11)
apical region (11-)13(-16) x (8-)9(-11)	7	(10-)13(-15) x (7-)9(-10)
	5	(-9)12(-14) x (8-)9(-10)
	3	(10-)12(-14) x (7-)8(-9)
	2	(14-)16(-18) x (9-)11(-13)

Tables 10, 11. – Cell sizes in µm of *E. radiata* plants no. 599 and 531 (wild material) and of 30 days old cultures isolated from these plants and kept at different salinities (7 = 34°/oo S; 6 = 25°/oo S; 5 = 17°/oo S; 4 = 9°/oo S; 3 = 4°/oo S; 2 = 1.5°/oo S).

– *E. TORTA* (Mert. in Jurg.) Reinb.

E. torta (Mertens in Jürgens) Reinbold 1893, p. 205; Bliding 1963, p. 41.

- Description (Figs 79-89)

Morphology (Figs 87-88)

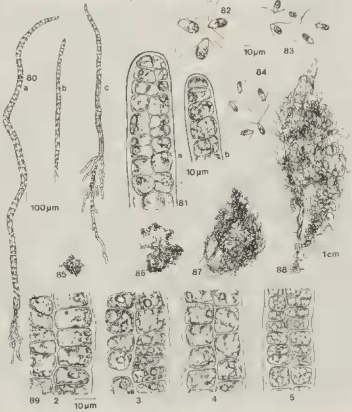
Thalli filiform, unbranched, of uniform narrowness, light to medium green. Stipe mostly absent in wild material, absolute length indefinable because of the twisted growth, but threads of about 50 cm can be isolated. Width of the thalli depending on the number of cell rows, visible in transection, from 30 to 45 µm in diameter; number of cell rows 4-10, central cavity 10-15 µm in diameter.

Anatomy (Fig. 79-81)

In wild material no distinction could be made between basal, middle and apical regions. In parts of the thalli with low division activity, the cells are more or less rectangular in longitudinal direction, and the cells are ordered in longitudinal rows; sometimes the cell rows have an oblique orientation in relation to the direction of the filament. In parts of the thalli with high division activity (young parts) the cells are more or less rectangular in transverse direction and

Figs 79-89. – *E. torta*. Fig. 79. two filaments, cells in surface view, plant no. 459. Fig. 80. germlings, a, c, after 7 days, b, after 5 days. Fig. 81. tips of filaments, a, without tip cell, b, with monoseriate apex. Fig. 82. zoospores. Fig. 83. female gametes. Fig. 84. male gametes. Figs 85, 86, 87, 88. 30 days old cultures from plant no. 600, Fig. 85 in 1.5°/oo S medium, Fig. 86 in 4°/oo S medium, Fig. 87 in 9°/oo S medium, Fig. 88 in 17°/oo S medium. Fig. 89. cells in surface view, same material as Figs 85-88. 2, 3, 4, 5, refer to media (see material and methods).

79 10 µm



the cell rows are relatively short, mostly consisting of 4-16 cells, separated from each other by thicker common cell walls; these cell rows show equal cell divisions. Cell walls 0.5-2 µm thick. In young parts the central thicker part of the parietal chloroplast, containing the pyrenoid(s), completely covering the outer cell wall. In older, slowly growing parts, the central thicker part of the chloroplast sometimes tilted towards one anticlinal cell wall, or the chloroplast only covering a limited part of the outer cell wall, with thin arms descending along the anticlinal cell walls. The chloroplast structure is seldom obscured by starch grains. Pyrenoids one per cell, two in about 5% of the cells, 2-4 µm in diameter, elliptic or round.

Reproductive cells (Figs 82-84)

In nature most filaments produce 2-flagellate gametes, the male ones smaller and with smaller chloroplasts than the female ones. They are positively phototactic, and are able to germinate, though sometimes in small numbers, without copulation. Sporophytic filaments produce big 4-flagellate zoospores, which are also positively phototactic.

Measurements, based on ten plants

cells in surface view	(12-)14(-19) x (6-)10(-14)	µm
– male gametes	(6-)6.5(-7.5) x (3.5-)4(-4.5)	»
female gametes	(7-)8(-9) x (5-)5.5(-6)	»
– zoospores	(9-)10.5(-12) x (5-)6(-7.5)	»

Morphology of germlings and young filaments (Fig. 80)

Zoospores and gametes germinate by forming a rhizoid, and immediately after that a strongly growing upright uniseriate filament. In a later stage these filaments grow into typical narrow cylinders, a central cavity is present when three cell rows are formed. The primary rhizoid branches in a later stage. The strongly growing filament never branches.

- Ecology and distribution

E. torta has been collected from five stations (see Table 12, and Fig. 1).

TABLE 12. Specimens investigated
(for locality numbers see Table 1, list of localities, in KOEMAN & van den HOEK, 1982)

2 (XII, '76, plants no. 611, 612); 2a (XII, '76, plants no. 615, 616); 5 (XI, '76, plants no. 600, 605, 609, 610); 6a (III, '76, plant no. 267); 19a (VI, '75, plant no. 206).

All stations were upper littoral mud or sandy mudflats, where the species was found growing in more or less sheltered places, loose lying in thin carpets, mostly entangled between other algae or phanerogams such as *Spartina* sp. and *Salicornia* sp. It was not found growing in non-marine habitats, though the results of culture experiments, which gave good growth in media with salinities ranging from 9-34°/oo S and poor growth in still lower salinities, do not exclude meso- to polyhaline stations as possible growing places (Figs 85-88).

- Morphological and anatomical characters in cultures (Figs 85-88, 89)

The important morphological and anatomical characters were retained in cultures. At salinities ranging from 25‰-9‰ S. the plants showed the same morphology as the original material. At salinities of 4‰ and 1.5‰ S the filiform plants were strongly twisted, which was caused by the abnormal expansion of some vegetative cells. Most cells contained one pyrenoid per cell and the chloroplast was mostly covering the outer cell wall. Plants cultured in low salinities had less uniform cell-dimensions, and cells were on average larger than those in high salinity media (Fig. 89; Table 13).

TABLE 13

wild material of *E. torta* 600	medium	cultures
various regions	5	(10-)11(-13) x (5-)7(-9)
	4	(11-)12(-14) x (7-)9(-12)
	3	(12-)14(-16) x (7-)9(-11)
	2	(15-)17(-18) x (9-)11(-13)

Table 13. – Cell sizes in μm of *E. torta* plant no. 600 (wild material) and of 30 days old cultures isolated from this plant and kept at different salinities (5 = 17‰ S; 4 = 9‰ S; 3 = 4‰ S; 2 = 1.5‰ S).

– *E. AHLNERIANA* Bliding

E. ahlneriana Bliding 1944, p. 338-345; 1963, p. 61-70

- Description (Figs 90-106)

Morphology (Figs 90-91)

Thalli strapshaped to filiform, the two layers compressed, in broad parts adnate with hollow margins, otherwise hollow, smooth or wrinkled, mostly not lubricous, light green to dark green, branched or seldom unbranched. Branched plants with branches of the first order along the whole main axis, the small ones more concentrated in the basal region of the thallus, the main ones more equally distributed along the axis. Main branches often branched, in the same way as the axis. Axis gradually narrowed towards the base into a fragile to firm stipe, depending on the dimensions of the plant, with a small disciform holdfast, from which new thalli may arise. Apices of axis and main (long) branches filiform to acute. Axis and main branches up to 1 m high and 2 cm broad, but mostly not more than 50 cm and 0.5 cm broad.

Anatomy, lower basal region (Figs 100, 102, 103)

In surface view, the stipe shows dark coloured rounded often elongate rhizoidal cells, which have about the same size as or are slightly bigger than the dark coloured vegetative cells. In this part cells form longitudinal rows. Cell

Figs 90-101. *E. ahlneriana*. Figs 90, 91. plants with zoids, plant no. 460 and 544. Fig. 92. branchlets, a, with long monoseriate apex, b, with one tip cell only, c, d, with short monoseriate apex. Figs 93, 94, 95. germlings, Figs 93, 94a after 5 days, Fig. 94b after 4 days, Fig. 95 after 7 days. Figs 96, 97, 98, 99. 30 days old cultures, same material as Fig. 90, Fig. 96 in 4°/oo S medium, Fig. 97 in 9°/oo S medium, Fig. 98 in 17°/oo S medium, Fig. 99 in 25°/oo S medium. Fig. 100. basis of plant no. 89. Fig. 101. cells in surface view, same material as Figs 96-99, B = basal region, M = middle region, A = apical region. 3, 4, 5, 6, refer to media (see material and methods).

Figs 102-106. — *E. ahlneriana*, cells in surface view, same material as Fig. 90. Fig. 102. lower basal region, lower zone. Fig. 103. lower basal region, upper zone. Fig. 104. upper basal region. Fig. 105. middle region. Fig. 106. apical region.

walls 3-6 μm thick. The central thicker part of the parietal chloroplast containing the pyrenoid(s) sometimes slightly tilted towards the apically oriented anticlinal cell wall or situated centrally against the peripheral cell wall. Descending arms relatively thin, descending along the anticlinal cell walls. The chloroplast structure, however, is mostly obscured by numerous small or medium sized starch grains. In surface view, the darker part just above the stipe shows many dark coloured smaller sized cells, whose contents have the same features as those in the lower zone; however, longitudinal cell rows are here more often disturbed by branching. Bases of young branches without rhizoidal cells. One pyrenoid per cell, two in less than 3% of the cells, 3-5 μm in diameter, round.

Anatomy, upper basal region (Fig. 104)

Cells in the upper basal region rounded, irregularly polygonal, or rectangular with rounded corners, showing nearly equal divisions, arranged in longitudinal rows, sometimes with an indication of transverse rows, but less ordered in densely branched parts. Cell walls 2-5 μm thick. The central thicker part of the parietal chloroplast containing the pyrenoid situated centrally against the peripheral cell wall or slightly tilted towards an anticlinal cell wall, with rather thin lobes descending along the anticlinal cell walls, or into the cell lumen. Some small starch grains may occur. One pyrenoid per cell, two in less than 2% of the cells, 2-4 μm in diameter, round.

Anatomy, middle and apical region (Figs 105, 106)

Cells in the middle and apical region rectangular, quadrangular or irregularly polygonal with or without rounded corners, showing equal divisions, arranged in longitudinal and short transverse rows, especially in broader parts of the thallus. Cell walls 0.5-2 μm thick, transverse ones on average thinner than the longitudinal ones. The central thicker part of the parietal chloroplast containing the pyrenoid situated centrally against the peripheral cell wall, with rather thin lobes, which may contain some small starch grains, descending along the anticlinal cell walls, or more seldom into the cell-lumen. One pyrenoid per cell, two in less than 2% of the cells, 2-3 μm in diameter, round.

Anatomy, tips of short filiform branchlets (Fig. 92)

In surface view, the monoseriate apex of young branches is mostly not more than three cells long. The tip cell is mostly slightly bigger than the other cells, which are on the whole somewhat smaller compared with normal thallus cells. Very young branchlets of about 3-6 cells are completely uniseriate, and without a thickened outer cell wall.

Reproductive cells (Figs 90, 91)

In the collected material only plants occurred which produced 4-flagellate zoids, or zoospores; the offspring of some of these plants produced 4-flagellate zoids again. They were always positively phototactic, and able to germinate very soon.

Measurements, based on nineteen plants

– axis, cells in surface view :		
lower basal region	(15-)18(-22) x (9-)12(-14)	µm
upper basal region lower zone	(12-)15(-18) x (9-)11(-12)	»
upper basal region upper zone	(10-)13(-15) x (7-)9(-11)	»
middle region	(10-)13(-15) x (7-)9(-10)	»
apical region	(11-)14(-16) x (8-)10(-12)	»
branches, cells in surface view :		
basal region	(12-)15(-19) x (10-)12(-14)	»
middle region	(12-)14(-17) x (9-)11(-12)	»
apical region	(9-)10(-12) x (8-)8(-9)	»
– zoids	(7-)8.5(-10) x (4.5-)6(-7)	»

Morphology of germlings and young fronds (Figs 93-95)

Zoids germinate by forming a short rhizoid, and immediately after that a strongly growing upright uniseriate filament. The first formed rhizoid branches densely; from some spherical rhizoid cells often more erect filaments are formed. At a later stage these filaments grow into hollow cylinders. The first formed main axis branches when growth continues, although less densely than in *E. ahlneriana* from nature, and it becomes compressed when large enough.

- Ecology and distribution

E. ahlneriana has been collected from nine stations (see Table 14 and Fig 1).

TABLE 14. – Specimens investigated
(for locality numbers see Table 1, list of localities, in KOEMAN & van den HOEK, 1982)

1b (VII, '75, plant no. 55); 3a (IV, '75, plant no. 16); 5 (V, '76, plants no. 473, 477, 479, 480; VI, '76, plants no. 540, 544); 6 (VI, '75, plant no. 89), 7a (V, '76, plants no. 458, 460; VI, '76, plants no. 505, 506, 517); 8 (VI, '75, plants no. 91, 98, 99); 22 (IV, '76, plant no. 332); 23 (IV, '76, plant no. 313).

Three of these stations were lower littoral and upper sublittoral zones on seadikes and harbour moles, exposed to strong wave action, where the species was growing attached to stones. Three localities were littoral mud or sandy mudflats, or sublittoral sediments in gullies among these mudflats, where the species was growing attached to mussels or other shells. Finally the species was encountered in three localities in meso- or polyhaline semi-stagnant or stagnant saline lakes. This accords with culture experiments which gave reasonable to good growth of germlings and young fronds in media with salinities ranging from 34-1.5‰ S (Figs 96-99).

- Morphological and anatomical characters in cultures (Figs 96-99, 101)

The important morphological and anatomical characters were retained in cultures, however, branching was less dense and in most cases more concentrated on the basal part of the stem. On the broadest thalli obtained from cultures with salinities of 17-25°/oo S some basally broad branches were developed higher up along the axis, as in most wild material. Most cells contained one pyrenoid per cell. The parietal chloroplast was completely covering the outer cell wall. Cells tended to be slightly smaller than in the wild material, and were generally ordered very well in longitudinal and transverse rows (see Table 15).

TABLE 15

wild material of *E. ahlneriana* 460	medium	cultures
upper basal region (10-)12(-14) x (7-)10(-12)	6	(7-)10(-12) x (6-)7(-7)
	5	(8-)9(-9) x (5-)6(-7)
	4	
	3	(12-)13(-15) x (8-)10(-12)
middle region (9-)12(-15) x (8-)9(-10)	6	(8-)10(-11) x (5-)7(-8)
	5	(6-)8(-10) x (5-)6(-6)
	4	(8-)9(-10) x (6-)7(-8)
	3	(10-)12(-14) x (8-)9(-10)
apical region (12-)16(-20) x (9-)11(-12)	6	(8-)10(-11) x (6-)8(-9)
	5	(10-)11(-13) x (6-)8(-9)
	4	(11-)12(-14) x (8-)9(-11)
	3	(13-)14(-16) x (8-)10(-13)

Table 15. — Cell sizes in μm of *E. ahlneriana* plant no. 460 (wild material) and of 30 days old cultures isolated from plant no. 460 and kept at four different salinities (6 = 25°/oo S; 5 = 17°/oo S; 4 = 9°/oo S; 3 = 4°/oo S).

DISCUSSION

The five species in the section *Proliferae* found in the Netherlands were also recognized by BLIDING (1963), namely *E. simplex* (as *E. prolifera* ssp. *prolifera* typus I), *E. prolifera* (as *E. prolifera* ssp. *prolifera* typus II), *E. radiata* (as *E. prolifera* ssp. *radiata*), *E. torta*, and *E. ahlneriana*. However, BLIDING ranged *E. torta* in his «Torta group», and not in his «Prolifera group» (which largely coincides with our section *Proliferae*). We range *E. torta* in the section *Proliferae* on the basis of its microscopic characters. Three more entities placed by Bliding in his «Prolifera group» were not found along the Dutch coasts, namely *E. prolifera* ssp. *prolifera* «Typus III» and «Typus IV», and *E. prolifera* ssp. *gullmariensis* Bliding.

In contrast to BLIDING (1963) we think that his three entities *E. prolifera* ssp. *prolifera* typus I, *E. prolifera* ssp. *prolifera* typus II, and *E. prolifera* ssp. *radiata* should be considered as three separate species. BLIDING's opinion is based on his claim that the three entities do interbreed and that intermediate forms do exist. However, interbreeding, especially under experimental conditions, indicates close affinity, but is no absolute criterium for conspecificity. With regard to morphological intermediates, one should realize that all taxonomically valid characters within *Enteromorpha* are of a quantitative nature, each character being represented by a graded series of variable expressions with overlaps between the species. Striking examples of species with such overlaps are *E. compressa*, *E. intestinalis* and three other species of the section *Enteromorpha* (KOEMAN & van den HOEK, 1982). Two arguments favour, in our opinion, the distinction of the three entities as separate species. In the first place these entities occur, in the field, as well recognizable populations in many different stations. *E. simplex*, for instance, is characterized in the first place by its (almost) simple fronds and its spirally twisted stipes. Populations of this species apparently occur in such widely distant places as the Netherlands (this work), Brittany (BLIDING, 1963), the White Sea, the Black Sea, the Bering Sea, and the Sea of Ochotsk (VINOGRADOVA, 1974, as *E. prolifera* f. *simplex* Vinogradova). In the second place the main characters are retained in unialgal cultures.

The species in the section *Proliferae* differ primarily from one another by their macroscopic morphology. This is also true for the section *Enteromorpha* (KOEMAN & van den HOEK, 1982). The sections, however, differ from one another mainly by microscopic anatomical characters. The main characters of the section *Proliferae* are the regular arrangement of the cells in longitudinal rows, the usual central position of the chloroplast against the peripheral cell wall, and the presence of mostly one pyrenoid per cell. On the basis of these characters *E. torta* has been included in the section *Proliferae*.

Three of the five species of the section *Proliferae* have wide ecological amplitudes with regard to exposure and salinity. *E. simplex*, *E. prolifera* and *E. ahlneriana* occur in the littoral and upper sublittoral zones of wave exposed to sheltered seadikes and harbour moles of euhaline to polyhaline tidal waters. They grow attached to solid substrates, such as loose shells, on sheltered intertidal mud and sandy mud flats. They are common in stagnant and semistagnant eu- to polyhaline man-made lakes in the southwestern Netherlands. *E. prolifera* may be even abundant in meso- and oligohaline canals and pools, where it grows attached to wooden poles and phanerogams, and where it may form loose floating masses. *E. simplex* cultures grow well at salinities of 9-34°/oo S, *E. prolifera* and *E. ahlneriana* cultures even at 1.5-34°/oo S.

E. radiata and *E. torta* have much narrower ecological ranges. Both species are limited to sheltered intertidal sand and sandy mudflats of euhaline to polyhaline tidal waters. *E. radiata* characteristically grows with its basal parts buried in the sediment, and it is only rarely attached to solid substrates; it extends from the mid littoral to the upper littoral zone where it may grow among salt marsh phanerogams (*Salicornia*, *Spartina*). *E. torta* mainly occurs in the latter habitat, where it forms a tangled mat on the surface of the sediment together with other saltmarsh algae, such as *Percursaria percusa*, *Enteromorpha radiata*, *E. ralfsii*, and *Rhizoclonium riparium* (cf. for instance NIENHUIS, 1970). Cultures of both species grow well at salinities of 9-34°/oo S.

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